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ABSTRACT

Behavioral measures, fractionated reaction and reflex times by means of electromyography, were used to determine if the eye color differences are found in the central or peripheral regions of the nervous system. The purpose of this research was to determine the truth of the hypothesis that dark-eyed individuals have faster reflex and reaction time than those with light eyes. Two experiments were conducted on subjects divided equally by eye pigmentation. The first fractionated simple reaction time into premotor and motor components by electromyography. Analysis of variance revealed that dark-eyed subjects had faster total reaction times and premotor times, but only the premotor time component approached significance. The second experiment fractionated patellar reflex time of light-eyed and dark-eyed subjects into reflex latency and motor components. There were no eye color differences for any of the reflex-time measures. The results of the two experiments support a central nervous system explanation for the iris pigmentation-reaction time phenomenon. (Authors/JD)

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Iris Pigmentation and Fractionated Reaction and Reflex Time

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Running Head: Iris Pigmentation

Iris Pigmentation and Fractionated Reaction and Reflex Time

Historically, science has had a symbolistic fascination for the eye. Natural and social scientists have recently begun to examine various parameters of the eye which are assumed to be readily observable indicators of underlying physiological and psychological processes. One such line of investigation (Worthy, 1974) proposes that iris pigmentation acts as an indicant of "reactive" perceptual-motor behavior -- a behavior characterized by "fast responses" under conditions that Poulton (1957) has termed "open skills" (e.g., the boxer reacting to the moves of his opponent). Worthy has stated that dark-eyed animals tend to excel in behaviors requiring speed, sensitivity, and reaction, while light-eyed animals tend to excel in behaviors requiring delay, self-pacing, or nonreaction.

There is a paucity of direct evidence for the effect of iris pigmentation on reactive motor behavior. Most empirical evidence, consisting of archival and case study comparisons, revealed that dark-eyed insects, rodents, and vertebrates have greater reactivity than do light-eyed strains within the same species (Worthy, 1974). More recent evidence (Landers, Obermeier, & Patterson, 1976; Wolf & Landers, 1978; Post, Note 1) has demonstrated that dark-eyed humans, regardless of race, sex, or socioeconomic status, consistently respond with faster reaction times (RTs) to auditory and visual stimuli than light-eyed

persons. This RT difference was only reliable between extreme pigmentation groups, with the magnitude of differences in the range of 10-23 msec.

The mechanisms suggested to underlie the proposed link between iris pigmentation and RT are related to the physical characteristics of melanin, a dark, granular pigment governing pigmentation in the skin, choroid, pia, and brain nuclei. Melanin in the eye is primarily located in the iris and to a lesser extent the cornea and sclera, and this melanin is qualitatively different from cutaneous, choroid, pia, and hair melanin (Kent, 1956). The iris is comprised of four melanized layers; it is the relationship of melanin in the more superficial layers to that in the posterior layers that determines eye coloration.

Because the eye is related embryologically to the motor control system in the central nervous system (CNS), it is highly probable that iris melanin and melanin in the motor system ("neuromelanin") are very similar and correlated to one another (Happy & Collins, 1972; Rolston, cited in Wasserman, 1974). Support for this assertion is provided by Bonaccorsi (1965), who found that iris pigmentation is positively correlated to the degree of melanization of neuroanatomical pathways in the inner ear. In addition, under certain conditions neuromelanin has been found to act as a semiconductor switching device which speeds the transmission of neural impulses (McGinness, Corry, & Proctor, 1974). If iris pigmentation and neuromelanin concentrations are highly correlated, darker iris pigmentation may indicate a greater capacity for afferent and/or

efferent neural transmission speed and efficiency, and enhanced reactive behavior. Iris pigmentation may also be genetically linked to the oxidative metabolism of catecholamines, such as dopamine (Edelstein, 1971), an inhibitory substance which when transmitted by the substantia nigra to the striated basal ganglia circuit assists in the central integration of motor activity. It may be that dark-eyed individuals produce a greater amount or faster turnover of dopamine, which could facilitate neural transmission. Presently, it is not known if the proposed mechanisms underlying eye color-RT differences are primarily central or peripheral nervous system manifestations.

The purpose of the experiments described here was to use behavioral measures, in this case fractionated reaction and reflex times by means of electromyography, to determine if the eye color differences are found in the central (premotor RT) or peripheral (motor RT and reflex times) regions of the nervous system. The premotor- and motor-time components of RT provide information relative to central and peripheral processing time, respectively (Weiss, 1965). The premotor time reflects the reception of the signal by the sense organ, the processing of the signal, and impulses sent from the brain to the corresponding muscles. The peripheral processing time, as indicated by motor time, is the duration from the onset of increased muscle activity to the desired response. Assuming that eye color affects central processing time, a fractionated RT should show brown- and blue-eyed group differences in the premotor component. If, on the other hand, the effect is assumed to be a peripheral phenomenon, the premotor RT should not differ, but the motor RT should show the eye-color effects.

Experiment 1

Method

Subjects

Right-handed male volunteers (N = 32) were selected from physical education basic instructional classes at The Pennsylvania State University on the basis of iris color as rated by two judges' comparisons with Worthy's 5-point descriptive categories (Worthy, Note 2), which were based on Kent's (1956) color plates showing shades of eye color. Sixteen light-eyed (range 1.0-1.9) and 16 dark-eyed (range 4.0-4.9) subjects were chosen.

Apparatus

The reactive movement was performed using a telegraph key mounted on a table. The key was wired to a Nicolet Digital Oscilloscope (Model #1090A), which recorded total RT. An electromyographical (EMG) trace recorded by two silver-silver chloride electrodes attached by electrode paste to the bulk of the extensor digitorum communis muscle of the right forearm delineated the premotor and motor components. A reference electrode attached to the back of the right hand reduced 60-cycle interference. A Healthkit IMP Scope was used to control the amplitude of the EMG muscle potential. The foreperiod interval and the light stimulus presentation were controlled with an interval timer. Subjects were separated from the measurement apparatus by a partition.

The light stimulus was located 30 in. (75 cm) in front of the subject in a 5 in. X 5 in. (12.5 cm X 12.5 cm) black box shielded on one side by opal glass. Located 1.5 in. (3.75 cm) and 4 in. (10 cm) in front of this box were a Kodak Wratten gelatin neutral density filter (No. 96, N.D. 2.00) and two 5 in. X 5 in. poloroid filters, respectively.

Procedure

At the beginning of the session subjects adapted to the semidarkness while the experimenter located the muscle bulk and attached the electrodes. The subject was then seated in front of and at eye level to the light stimulus with his chin on the rest and right index finger on the reaction key. The elbow angle was positioned at 90^{o}. To adjust for individual differences in perceived brightness individual threshold values were found by rotating the proximal polaroid filter slowly to the right to the location where the subject could first detect the stimulus. The neutral density filter was then removed, increasing the intensity by 2 log units over the individual's threshold value.

The subject received 13 practice trials (which included three catch trials) and 21 performance trials. Each trial was preceded by a set of commands -- "depress the key" and "ready" -- and a randomized foreperiod of 2, 3, and 4 sec. Knowledge of results in msec for total RT was given after each practice trial, but not for performance trials. An intertrial-rest period of approximately 15 sec followed each performance trial.

Results and Discussion

Subjects' light threshold levels were compared for the two iris pigmentation groups (dark-eyed $\underline{M}$ = 14.81 and light-eyed $\underline{M}$ = 12.81). These means were not significantly different, $\underline{t}$ (30) = 1.49, $\underline{p} > .05$, which suggests that the origin of the eye color-motor reactivity difference is not a function of subjects' subjective perception and sensitivity to brightness.

The data for premotor, motor, and total RT were blocked into three blocks of seven trials. The means and the standard deviations for each eye color group are shown in Table 1. Dark-eyed subjects had shorter latencies in both the premotor and total RT measures than the light-eyed subjects. Motor times were similar, but the light-eyed subjects had faster times than the dark-eyed subjects. An eye color by blocks (2 X 3) repeated measures ANOVA showed no significant differences between the two eye color groups on any of the behavioral measures. However, for the premotor component, the eye-color main effect approached significance, $\underline{F}$ (1, 30) = 3.53, $\underline{p} < .07$, which was considered as supporting Worthy's (1974) reactivity hypothesis. The nonsignificant eye color effect for total RT was in the predicted direction, which was the result of differences between eye-color groups in the premotor phase.

Insert Table 1 about here

These mean differences in RT were smaller than those obtained in previous studies (Landers et al., 1976, Experiment 2; Landers, Obermeier, & Wolf, 1977; Wolf & Landers, 1978). In the present investigation a 10.5 msec difference for premotor time and 8.5 msec difference for total RT was obtained. In the other experiments, the total RT advantage of dark-eyed groups averaged 18 msec. The somewhat weaker statistical results reported for this study may have been due to the smaller number of subjects (N = 32), which was one-third less than in previous studies.

The blocks main effect for total RT and premotor time were highly significant indicating improvement over trial blocks, F (2, 60) = 6.61, $p < .003$, and F (2, 60) = 5.46, $p < .007$, respectively. The interactions of groups by blocks were nonsignificant for all three dependent variables. It appears that subjects were reacting faster as the trials progressed, and an expected learning or practice effect had occurred.

According to our initial hypothesis, the mean differences in the premotor phase support a central origin of the eye color phenomenon. Although these results clearly show that the effect is not in the motor phase, it does not negate the implication of other peripheral influences. Therefore, another experiment was undertaken which fractionated a simple monosynaptic reflex into reflex latency and motor components in order to determine if the effect originates in peripheral afferent and/or efferent nerves or the CNS. Nonsignificant differences in both reflex latency (peripheral afferent and efferent conduction) and motor (muscle contraction) components of a monosynaptic response would offer additional evidence that the locus of eye color-RT differences resides in the CNS.

Experiment 2

Method

Subjects

Subjects were male volunteers (N = 30) selected from The Pennsylvania State University faculty and student body on the basis of Worthy's (Note 2) iris pigmentation categories. Half were light-eyed (range 1.1-1.5) and half were dark-eyed (range 4.2-4.8).

Apparatus

The apparatus employed in this study has been described in a previous paper (Wolf & Landers, 1978). The base of a Lafayette Patellar Reflex Apparatus (Model 78025) rested on a table. The vertical frame housing the pendulum hammer assembly and the rotating shaft to which it was attached was mounted directly to the right of the subject along the right upper thigh. The hammer assembly (weight 400 g) consisted of a microswitch which made contact with the right patellar tendon simultaneously with the impact of the rubber hammer. Impact closed the microswitch and actuated an oscilloscope. The hammer assembly was redesigned so that it was in a fixed position at the end of the pendulum shaft, all vertical adjustments being made at the pivot point on the rotating shaft.

A second apparatus employed a switch box which was adjustable in all three planes of movement. The box was positioned for each subject at 3.5 in (9 cm) directly posterior to the malleoi of the ankle. A U-bolt assembly was fastened just above the malleoli which had a 3 in. (7.5 cm) bolt that inserted into the top frame of the switch box and held the circuit open by keeping the spring loaded plunger-switch

depressed. Upon initiation of ankle movement, the pin was pulled out releasing the plunger, closing the circuit, and stopping the oscilloscope impulse. Total reflex time was measured as the interval between hammer strike and initiation of ankle movement.

The angle from which the hammer was released was measured by a protractor mounted on the verticle frame. To release the hammer, the experimenter depressed a switch which deactivated an electromagnet. The angle of release was held constant at suprathreshold level for each subject, defined as 10° above individually determined threshold levels.

Total reflex time was fractionated into reflex latency and motor components by EMG. Two silver-silver chloride electrodes were attached approximately 1 in. (2.54 cm) apart over the prominence of the rectus femoris muscle on the right thigh approximately midway between the hip joint and knee joint. A reference electrode was attached on the back of the right hand. A Tektronix (Type 122) Low Level Preamplifier amplified the extensor muscle contraction resulting from the elicited patellar tendon reflex, and a Tektronix (Model 5103 N) four-channel oscilloscope allowed the investigator to visually identify and record in msec the stimulus initiation (hammer switch), initial muscle contraction, and movement switch closure.

Since the patellar tendon reflex is classified as a monosynaptic stretch reflex, reflex latency has been defined (Hayes, 1972) as "representing the sum of neuronal components of the reflex arc." Motor time

was defined (Hayes, 1972) as "the time taken for the spread of excitation and development of sufficient tension within the muscle(s) to overcome inertia and set the limb in motion." Total reflex time combines reflex latency and motor components.

Procedure

Subjects donned athletic shorts and removed their shoes upon arrival at the laboratory. They extended the right leg so the muscle bulk could be located, then the areas on thigh and hand were abraided, cleaned, and electrodes were attached and connected to the preamplifier.

With subjects seated on the base of the reflex apparatus, wooden spacers were placed outside the lateral surface of the thigh, and a velcro strip was fastened around the thigh to the frame to keep the leg in place. The hammer assembly was positioned so that contact would be made with the patellar tendon just distal to the patella. The switch box was positioned directly behind and above the ankle malleoli so the pin could move freely outwards when pulled by ankle movement. Threshold for reflex elicitation was defined as the angle at which 60% of the strikes would elicit release of the depressed plunger-switch. The angle of release was increased in small increments from an initial angle of 30° from the patella until threshold was reached.

Subjects were instructed to keep muscles relaxed, but remain mentally alert and keep their eyes closed during testing. White noise was presented through a pair of headphones to prevent auditory cues and anticipation of the hammer strike. Visual inspection of the oscilloscope

grid allowed the experimenter to judge if muscle contraction had occurred prior to hammer strike. Twenty hammer strikes were administered to each subject with an intertrial-rest period of 15-30 sec.

Results and Discussion

The data for reflex latency, motor, and total reflex times were blocked into four blocks of five consecutive trials. These means and standard deviations for each group are contained in Table 2. The data were then analyzed in three separate 2 X 4 (eye color X blocks) ANOVAs. The main effect for eye color and the eye color X blocks interaction were nonsignificant for all three measures ($Fs < 1.45$). It may be noted that reflex latency means are slightly larger than the range reported in previous studies, but comparison with reported standard deviations (Hayes, 1972; Kroll, 1968; Knowlton & Britt, 1963) indicates the present values to be within the normal range. This suggests that the group means were inflated slightly by some constant amount, probably due to differences in timing stimulus onset and the termination of the response.

Insert Table 2 about here

These results replicate the total reflex-time results of Wolf and Landers (1978), and together they indicate that melanization of peripheral afferent and efferent nerves may not produce eye color-related differences in involuntary, reflexive responses. Although it would be dangerous to conclude on the basis of a single study that the null hypothesis is

supported, thus far the cumulative evidence shows eye color differences in RTs which are consistent with a CNS mechanism.

The blocks main effect was significant for motor time, F (3, 84) = 4.43, $p < .05$, marginally significant for total reflex time, F (3,84) = 4.01, $p = .055$, and nonsignificant for reflex latency, F (3, 84) = 1.69, $p > .05$. Examination of block means for motor time and total reflex time indicated that subjects' last three blocks were approximately 8 and 7 msec slower respectively than the first block. This finding replicates Wolf and Landers (1978) finding. Jendrassik (1883) originally reported that any prior muscle tension before reflex elicitation will facilitate the magnitude and possibly the speed of the reflex. Subjects may have been initially aroused and then gradually relaxed as testing proceeded. This phenomenon could also be explained by progressive stimulus habituation in increasing numbers of unresponsive stretch receptors evoked by repeated hammer strikes to the patellar tendon. Several subjects did mention experiencing this latter effect.

General Discussion

The results of the two investigations clearly suggest two conclusions. First, the cumulative evidence (Landers et al., 1976; Wolf & Landers, 1978; Post, Note 1) indicates that dark-eyed individuals consistently respond with significantly faster RTs to auditory or visual stimuli than light-eyed persons. Since nonsignificant differences

between eye-color groups has been reported in choice response time and simple response and movement time (Landers et al., 1976; Wolf & Landers, 1978), Worthy's (1974) reactivity hypothesis appears to be limited to a strict definition of the RT component of various motor skills.

Although the predicted eye color differences in premotor RT were only marginally significant in the first experiment, this lack of statistical significance perhaps resulted from a reduced subject population. It is important to bear in mind that even though the sample size and statistical probability levels may vary from study to study, the differences in premotor-time and simple RT have always favored dark-eyed subjects. Considering that Worthy's hypothesis has been experimentally tested five times with five different samples (Landers et al., 1976, Experiment 2; Landers et al, 1977; Landers & Wolf, 1978; Post, Note 1; and Experiment 1 of this study), a combined probability value would more accurately reflect the reliability of the eye color phenomenon. This is based on the premise that if replications are of value, there must be some way in which the results of replications have some cumulative bearing on the nature of some relationship. Therefore, using a _z_-transformation procedure suggested by Mosteller and Bush (1954), a _z_ value was obtained that could not occur by chance any more than three times in 10 million (Table 3). Worthy's hypothesis, therefore, reliably predicts RT differences between eye color groups from one study to the next.

Insert Table 3 about here

The second conclusion suggested by the data is that the replicated findings of eye color differences in RT seem to be produced by a CNS mechanism possibly associated with the degree of neuromelanin distribution. The nonsignificant (total) patellar reflex differences reported by Wolf & Landers (1978) and Experiment 2 of this study suggest that the locus of causation is not found in peripheral neural circuits. Furthermore, the nonsignificant motor-time differences in Experiments 1 and 2 clearly discount the differentiation of muscle fiber type (fast twitch vs. slow twitch) as a viable explanation.

Since eye color is a genetic, sex-linked trait carried on the X chromosome (Brues, 1946), it seems probable that pigmentation in the CNS is of the same genetic origin. Although no direct evidence has as yet established a link between iris color and neuromelanin in the CNS, neuromelanin is heavily concentrated in CNS locations that are important for motor control. Bazelon, Fenichel and Randall (1967) identified a column of melanized cells extending the entire length of the human brainstem (midbrain to the cervical spinal cord) which closely follows the course of the general visceral efferent column of the brainstem. The most heavily melanized brainstem nucleus, the substantia nigra (SN), is closely interconnected with the basal ganglia and thalamic nuclei, and is an integral part of several extrapyrimidal motor circuits and feedback loops (Clark, 1975). Happy and Collins (1972) have also noted a pigmentation of the ascending reticular formation. Thus, the SN has connections, direct or indirect, to most structures of the CNS involved in motor control.

According to one explanation (Worthy, 1974), if iris pigmentation and neuromelanin concentrations are highly correlated, the semiconductor properties of neuromelanin may allow dark-eyed individuals to have increased afferent and/or efferent neural transmission speed and efficiency. Recent evidence by Borg (1974), however, casts doubt on the plausibility of this explanation since the free radical signals in melanins represent only 1% of the units and seem to be randomly scattered. He also cites Boyce's conclusions that melanins are really not semiconductors since the free radicals are trapped by surrounding electrons.

Although the semiconductor (threshold switching) explanation is controversial, neuromelanin may function in other ways. Forest (1974) views neuromelanin as constituting an important factor in the capacity of the pigmented brain nuclei to arouse and excite higher brain centers, including the limbic system and other cortical regions which facilitate consciousness. Thus greater neuromelanin concentrations may indicate a more "reactive" nervous system manifesting itself in various forms, one being enhanced psychomotor stimulability. This is consistent with evidence showing that the absence of neuromelanin in several neural structures (e.g., SN) is characteristic of patients with diseases such as Parkinson's, Intention Tremor, and Mongolism, which are associated with defective motor control symptoms (Kent, 1956). For example, Parkinsonian patients have slower RTs than normal subjects (Brumlik & Boshes, 1966; Heilman, Bowers, Watson, & Greer, 1976). In addition,

Horvath and Meares (1974) have found a significant reduction in premotor RT after the administration of L-dopa to Parkinsonian patients. This implicates dopamine concentration as a possible underlying mechanism for the iris color-motor reactivity relationship.

Dopamine, one of the three naturally occurring catecholamines, seems most important in that mechanisms underlying the central integration of motor activity are closely related to the dopamine levels in the midbrain (Bazelon, 1967; Ericson, 1971). Iris color may be genetically linked to the neural metabolism of dopamine, which is converted to neuromelanin by enzymatic action (Bazelon et al., 1967; Wasserman, 1974). Iris color, therefore, may be an indicant of neural metabolism of catecholamines, rate of receptor binding of dopamine, or the number of postsynaptic receptors for this neurotransmitter. If it is an indicant, greater iris pigmentation would result in high levels of these factors, leading to faster impulse transmission.

Although not mutually exclusive with the previous explanation, another possibility for faster neural transmission is a form of lateral inhibition. In voluntary movement, the cerebellum and the basal ganglia are activated at almost exactly the same time as the motor cortex. These subcortical areas maintain a postural background for voluntary movement. The SN has been directly implicated in the initiation of movement (Carlsson, 1972). On the other hand, the caudate nucleus, which is connected to the SN through nigrostriatal fibers, has been directly implicated in the inhibition (through the nonspecific system to the cortex) of all non-essential sensory information. Perhaps these inhibitory fibers provide

a sort of inhibitional tuning or attentional filtering so as to allow the primary motor signal to dominate. Thus, those with greater inhibitory power may be able to respond faster to stimuli. Neuromelanin may influence this system through its relationship with dopamine, the primary inhibitory neurotransmitter in these structures.

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Table 1

Means and Standard Deviations of Premotor, Motor, and Total Reaction Time (in msec) of Light- and Dark-eyed Subjects

Group	Premotor		Motor		Total	
	Mean	SD	Mean	SD	Mean	SD
Dark-eyed	201.99	17.06	50.69	6.67	252.69	19.26
Light-eyed	212.62	18.33	48.52	6.37	261.14	20.68

Table 2

Means and Standard Deviations of Reflex Latency, Motor, and Total Reflex Time (in msec) of Light- and Dark-eyed Subjects

Group	Reflex Latency		Motor		Total	
	Mean	SD	Mean	SD	Mean	SD
Dark-eyed	31.23	1.22	78.57	10.21	109.93	10.82
Light-eyed	29.41	1.33	82.56	11.08	111.98	11.49

Table 3

Reaction Time Differences of a Series of

Replications Utilized to Computer a Cumulative Probability

Study	P (One-Tailed)	(N)	z	Average advantage of Dark-eyed over Light-eyed
Landers et al., 1976, Exp. 2	.04	(48)F	1.750	+.230
Landers et al., 1977	.064	(70)r_s	1.610	+.207
Wolf & Landers, 1978	.001	(60)F	3.100	+.120
Exp. 1, Present Study	.07	(32)F	1.475	+.106
Post, Note 1	.0003	(620)F	3.400	+.101

Sum z = 11.34 $z = \text{Sum } z / \sqrt{N\ 5} = 5.08$

Two-tailed $p < .0000003$